

METHODS FOR GENERATING DOUBLED HAPLOID PLANTS

Abstract of the Disclosure

The present invention provides methods for generating doubled haploid and/or haploid plants from microspores. In a presently preferred embodiment of the methods of the present invention, plant material is selected that bears reproductive organs containing microspores at a developmental stage that is amenable to androgenic induction. The microspores are treated by contacting the selected plant material with water and subjecting the selected plant material to temperature stress, and optionally to nutrient stress. Preferably the selected plant material is contacted with an effective amount of a sporophytic development inducer and an effective amount of an auxin and/or a cell spindle inhibiting agent. Optionally, the selected plant material is contacted with an effective amount of a cytokinin and/or an effective amount of a gibberellin. The treated microspores are isolated, preferably by density centrifugation utilizing a solution of 0.3 M mannitol layered over a higher density solution of a sugar, preferably maltose. The isolated, treated microspores are then cultured in a liquid nutrient suspension medium supplemented with at least one plant ovary or with an aliquot of plant ovary conditioned medium, until the microspores develop into embryoids. The embryoids are transferred to a regeneration medium and incubated therein until the embryoids develop into plants. The resulting plants may be haploid or doubled haploid and may also be genetically transformed.